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Assessment of occurrence, diversity, and biomass of macroinvertebrates in Swiss groundwater systems using citizen science data

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Abstract

Groundwater is a vast ecosystem harboring a high diversity of specialized taxa. Despite its diversity, groundwater is a still relatively unexplored and threatened ecosystem. Especially the linkage of groundwater with other ecosystems remains largely unknown from the perspective of groundwater fauna. Here, we used citizen science data to get a first baseline knowledge of the occurrence, diversity, and biomass of major macroinvertebrate groups found in shallow groundwater systems of Switzerland. We investigated all organisms collected from the groundwater in 346 spring catchment boxes of municipal drinking water providers. We morphologically identified the organisms on a broad taxonomic level and estimated their biomass and pigmentation using automated image processing. Crustaceans, particularly *Niphargus* and groundwater isopods, were the most common taxa of obligate groundwater organisms. We also found a surprisingly high number of macroinvertebrates associated with surface and subsurface ecosystems. These taxa might be accidentally entering the groundwater or use it as temporary habitat. In both cases they possibly contribute essential allochthonous energy imports from the surface. We found a positive relationship between the estimated biomass of macroinvertebrates in the samples and the occurrence and abundance of *Niphargus*. Owing to the widespread occurrences of surface and subsurface macroinvertebrates in our groundwater samples, our study provides evidence for common interactions between groundwater, soil, and surface ecosystems.

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Keywords
Aquifers, hyporheic, pigmentation, stygofauna, subsidy, Switzerland

Introduction

Groundwater harbors a unique and diverse fauna, yet is still an understudied ecosystem (Mammola et al. 2020). This stygofauna contributes substantially to the functioning of groundwater ecosystems, for example by facilitating the breakdown of particulate organic matter (Boulton et al. 2008; Griebler and Avramov 2015). It consists of obligate groundwater organisms (stygobites), as well as organisms that are occasionally or accidentally entering the groundwater realm (stygophiles and stygoxenes) (Gibert et al. 1994). Many of these organisms are vulnerable to rapid environmental changes, making them essential conservation subjects and potential biological indicators for monitoring groundwater quality (Malard et al. 1996; Griebler et al. 2010; Stein et al. 2010).

While attempts to develop ecological indicators for groundwater monitoring have been made (e.g., Hahn 2006; Steube et al. 2009; Korbel and Hose 2011; Griebler et al. 2014a), there is still a long way to go until this method becomes widely applied (Steube et al. 2009; Griebler et al. 2014a). This also reflects the state of research on groundwater ecosystems, which lags behind that of surface water ecosystems (Danielopol et al. 2003; Griebler et al. 2014b; Borko et al. 2022). One of the main causes is the restricted accessibility to groundwater ecosystems (Gibert and Culver 2009; Griebler et al. 2014b). In addition, ecological links between groundwater and other ecosystems have been understudied, despite groundwater aquifers being inherently linked to surface water and soil through water flows and groundwater recharge (Malard et al. 2023).

Along the hyporheic zone, water, dissolved oxygen, nutrients, organic matter, and organisms are exchanged between surface water and groundwater, creating environmental gradients along this transition area (Boulton et al. 2008). Since aquifers are environments that lack photosynthetic primary production, groundwater fauna largely relies on allochthonous energy sources imported from the surface (e.g., Gibert et al. 1994; Humphreys 2006; Fišer et al. 2014), making it a typical example of resource subsidies and meta-ecosystem dynamics (see Gounand et al. 2018). In specific cases, this is complemented by chemolithoautotrophic primary production within groundwater ecosystems themselves (Hutchins et al. 2016; Herrmann et al. 2020). An essential contributor to the energy flow from the surface to groundwater ecosystems is the total inflow of macroinvertebrate biomass (e.g., Benke and Huryn 2007; Machuca-Sepúlveda et al. 2022; Malard et al. 2023).

Drinking water aquifers and subsequent water collection in spring catchment boxes provide novel yet underexplored access to groundwater systems (Alther et al. 2021; Couton et al. 2023a, 2023b). Due to the shallow depth from which the groundwater is drained from the aquifers into these facilities, they enable the study of groundwater biodiversity and the linkage of groundwater to other ecosystems. In Switzerland, promising monitoring approaches of these spring catchment boxes have been estab-

lished (e.g., Alther et al. 2021; Couton et al. 2023a). For example, studies by Alther (2020) and Studer et al. (2022) show that a citizen science approach is successful in accessing groundwater ecosystems through spring catchment boxes. However, these studies have focused mainly on groundwater amphipods (*Niphargus* spp.) and specific regions of Switzerland, and therefore, the entirety of groundwater fauna across all of Switzerland and the interactions between groundwater and other ecosystems still remain largely unknown and undocumented.

Here, we shed light on the diversity, occurrence, and biomass of macroinvertebrates found in Swiss groundwater systems and on the linkage to other surface and subsurface ecosystems. We used standardized groundwater samples retrieved from spring catchment boxes by local drinking water providers as part of a systematic Swiss-wide citizen science project. We measured the biomass of macroinvertebrates collected in groundwater to understand ecological processes between groundwater and surface ecosystems and its potential as resource influx. Additionally, we used the pigmentation of the macroinvertebrates as an approximate classification into hypogean and epigean fauna.

Methods

Citizen science sampling procedure

The samples were collected between 2021 and 2022 using a Swiss-wide, systematic citizen science approach. We collaborated with local drinking water providers, who sampled the groundwater flowing into spring catchment boxes (hereafter referred to as spring boxes) with filter nets (similar to Alther et al. 2021 and Studer et al. 2022). First, we used a regular grid to select municipalities and contacted the corresponding drinking water providers to ask for participation in our study. We then sent sampling kits and sampling instructions to all drinking water providers interested in participation. The water providers were instructed to install a filter net (mesh size 0.8 mm, Sefiltec AG, Höri, Switzerland) around pipes that passively drain groundwater from the aquifer to the spring box and to repeatedly collect all organisms washed into the filter net every seven days. All organisms were transferred into sample tubes containing 80% ethanol. The samples and a protocol containing supplementary information such as sampling duration and water discharge rate were then returned to us. After receiving the samples, we separated amphipods from other macroinvertebrates and stored all samples at 4 °C. For the subsequent analysis, we included 1,182 samples from 346 sites across Switzerland, for all of which data on water discharge rate and standardized sampling duration was available.

Taxonomic identification

All macroinvertebrates were identified morphologically using a stereomicroscope (Nikon SMZ1500, 0.75–11.25×). We also identified exuviae and fragments of animals when the number of individuals could be inferred. Based on various identification

resources (Zettel 2003; Tachet et al. 2006; Schminke and Gad 2007; Altermatt et al. 2019; Klausnitzer 2019; Zaenker et al. 2020; Walser et al. 2021), specimens belonging to the classes Symphyla, Chilopoda, Gastropoda, and Diplopoda were identified to class level, while Hirudinea, Oligochaeta, Collembola, Acari, and Diplura were identified to subclass level. Specimens of the following taxa were identified to order level: Ephemeroptera (larvae), Plecoptera (larvae), Trichoptera (larvae), Coleoptera (adults and larvae), Hemiptera, Diptera (larvae), Isopoda, Araneae, Opiliones, and Pseudoscorpiones. Formicidae (Hymenoptera) specimens were identified to family level, and Amphipoda specimens to genus level. For the order Isopoda, we differentiated between groundwater Isopoda (Asellidae, cf. *Proasellus,* unpigmented) and remaining Isopoda (pigmented). All other specimens not belonging (or not identifiable) to any of these groups were summarized as "unidentified" or "other adult insects" respectively.

Estimation of biomass and pigmentation

We used automated image processing (ImageJ version 1.53t, Rasband 2014) to identify the silhouette of the organisms within each sample and then calculated the area values and the mean grey values (as an estimate of their pigmentation level) of the respective silhouettes. For each sample, we used the sum of the area values (hereafter referred to as bio-area) as a proxy for biomass, as we found a strong positive correlation between the two in a subset of the data (Suppl. material 1). To create the images, we transferred all macroinvertebrate specimens (including fragments and exuviae) of each sample into a petri dish (d = 8.5 cm) filled with 80% ethanol. We placed the petri dish in a predefined position over a blue, laminated paper that contrasted with both pigmented and unpigmented specimens (Suppl. material 2: fig. S2). We took one picture of each sample with a digital camera (Nikon D5600 with Nikkor 18–55 mm lens) fixed to a camera stand. We measured the area (mm^2) of each organism and quantified its average pixel intensity with batch processing in ImageJ (see Suppl. material 2 for batch processing code and additional information). The CSV files containing the area measurements and the mean grey values of all individuals of a sample were processed in R version 4.1.2 (R Core Team 2021). Measurements for *Niphargus*, *Gammarus*, and oligochaetes were compiled and added separately, as these specimens were previously sorted out for further work (Suppl. material 2).

Statistical analysis

We standardized the bio-area and taxonomic abundances by the sampling duration and the volume of discharged groundwater (retrieved from the sampling protocol filled by the drinking water providers). Therefore, we calculated the bio-area of each site per 100 megaliters of discharged groundwater (1 ML = 1,000,000 liters). For the taxonomic abundances we standardized by 1,000 ML discharged groundwater per site. Based on the mean grey value of all organisms, we additionally split the standardized bio-area of each sample into light-pigmented (mean grey value above 111.86) and

dark-pigmented (mean grey value below 111.86) bio-area. The threshold for the categorization was set based on the mean grey values of groundwater Isopoda (Asellidae, cf. *Proasellus*, unpigmented) specimens. All statistical analyses were performed based on the standardized data.

We extracted for each sampling site the aquifer type and tested if diversity, bio-area and pigmentation ratio varied between aquifer types. Therefore, we included the three dominant aquifer types present in Switzerland, namely fissured, karstic, and unconsolidated aquifers. The geodata for the aquifers was retrieved from BAFU (2017). We calculated the following local diversity metrics based on taxonomic orders for each site: richness, Shannon index, and Pielou's evenness. These indices were calculated using the R package vegan (version 2.5–7, Oksanen et al. 2020). We then evaluated whether the diversity metrics, as well as the total standardized bio-area and proportion of light-pigmented bio-area values differed between aquifer types using Kruskal-Wallis rank-sum tests and post-hoc pairwise Wilcoxon rank-sum tests with Bonferroni correction.

To test for a difference between the amounts of light- and dark-pigmented, standardized bio-area per sample, we used a paired Wilcoxon rank-sum test. For samples including both pigmentation categories, we additionally computed Kendall's Tau to assess the correlation between the amounts of light- and dark-pigmented bio-area of the samples. We analyzed the effect of the bio-area on *Niphargus* occurrence with a generalized linear model (GLM), using a binomial error distribution. Twelve sites with a very high amount of bio-area (> 40,000 mm^2 per 100 ML discharged groundwater) had a large impact on the results of the model and were thus removed from the analysis. To analyse the relationship between *Niphargus* abundance and macroinvertebrate bio-area, we compared three different models, that all accounted for zero-inflation, since *Niphargus* was not detected in 66.18% of the sampling sites. We applied two zero-inflated negative binomial (ZINB) models (with and without square-root-transformed response and explanatory variables) and a zero-inflated Poisson (ZIP) model, using the R function "zeroinfl()" from the package pscl (Jackman 2020). The best model was selected based on the dispersion statistic and the Akaike Information Criterion (AIC).

All analyses were performed using RStudio version 2023.03.0+386 on R version 4.1.2 (R Core Team 2021). Figures were produced using the package ggplot2 (Wickham 2016).

Results

Using a citizen science approach, we obtained 1,182 standardized filter net samples collected by municipal drinking water providers from 346 spring boxes (Fig. 1A). For all of these samples, data on water discharge rate and sampling duration was available.

Overall, 404 samples were empty and 778 samples contained a total of 5,578 macroinvertebrate individuals (including fragments and exuviae). Out of those, we identified 5,390 individuals belonging to 9 classes (Insecta, Malacostraca, Diplopoda, Chilopoda, Symphyla, Arachnida, Clitellata, Gastropoda, and Entognatha). 4,408 of

those individuals were additionally identified to the order level. The remaining 188 individuals could not be identified.

Plecoptera (larva) and the two stygobiotic taxa *Niphargus* and groundwater Isopoda (Asellidae, cf. *Proasellus*, unpigmented) were the most abundant taxonomic groups. The least abundant taxon was Diplura (Suppl. material 3: fig. S4). We found macroinvertebrates in 271 out of 346 sites (78%) (Fig. 1A). The three taxa found in most spring boxes were *Niphargus* (34% of sites), other adult Insecta (30% of sites), and adult Coleoptera (27% of sites) (Fig. 1B). Groundwater Isopoda (Asellidae, cf. *Proasellus*, unpigmented) were found in 21% of sites (Fig. 1B).

When comparing the local macroinvertebrate diversity (Shannon index) and richness at the order level between aquifer types, a Kruskal-Wallis test revealed a statistically significant difference in local diversity ($X^2_{(2)} = 6.54$, $p = 0.038$) and richness ($X^2_{(2)} = 15.04$, $p < 0.001$) (Fig. 2). Post-hoc pairwise Wilcoxon tests indicated a higher median for diversity in unconsolidated aquifers in comparison to fissured aquifers ($p < 0.05$). For richness, both, unconsolidated ($p < 0.05$) and karstic ($p < 0.05$) aquifers had higher medians in comparison to sites within fissured aquifers. Despite finding some evidence for a difference in Pielou's Evenness ($X^2_{(2)} = 6.02$, $p = 0.049$), none of the pairwise comparisons between aquifer types was significant.

The amount of standardized macroinvertebrate bio-area ranged from 0 to 396,991 mm^2 per 100 ML discharged groundwater. The median was 953 mm^2 per 100 ML discharged groundwater. Using a Kruskal-Wallis test, we did not find any significant difference of the standardized bio-area between different aquifer types ($X^2_{(2)} = 4.56$, $p = 0.10$).

In total, 40% of the standardized macroinvertebrate bio-area sampled across all Switzerland was light-pigmented and 60% dark-pigmented. Based on the 271 sites where any bio-area was obtained, 67% of the sites had dark and light-pigmented bio-area, whereas 20% had dark-pigmented only and 13% light-pigmented bio-area only. The median amount of dark-pigmented standardized bio-area per sample was significantly higher than the median amount of light-pigmented area (paired Wilcoxon test, $p < 0.05$, Fig. 3). For samples including both pigmentation categories, we found a positive correlation between the amounts of standardized light- and dark-pigmented bio-area (Kendall's Tau = 0.48, $p < 0.05$). No significant difference was found when comparing the proportions of light-pigmented bio-area per sample between aquifer types (Kruskal-Wallis test, $X^2_{(2)} = 0.35$, $p = 0.84$).

Based on the standardized data, we compared the occurrence and abundance of *Niphargus* with the bio-area of other macroinvertebrates. There were 75 sites with empty samples (no *Niphargus* and no other macroinvertebrate bio-area) and 105 sites where both *Niphargus* and other macroinvertebrates were obtained. There were 154 sites without *Niphargus* but with other macroinvertebrates (bio-area of other macroinvertebrates > 0) and 12 sites where only *Niphargus* was detected (bio-area of other macroinvertebrates = 0). The binomial GLM showed a tendency for *Niphargus* occurrence to increase with increasing bio-area of other macroinvertebrates (Fig. 4 and Suppl. material 3: table S1). Based on the dispersion statistic and the AIC, we selected the ZINB model with square-root-transformed response and explanatory variables (Suppl.

material 3: table S2). The ZINB regression model showed that *Niphargus* abundance significantly increased with increasing bio-area of other macroinvertebrates (Fig. 5 and Suppl. material 3: table S3).

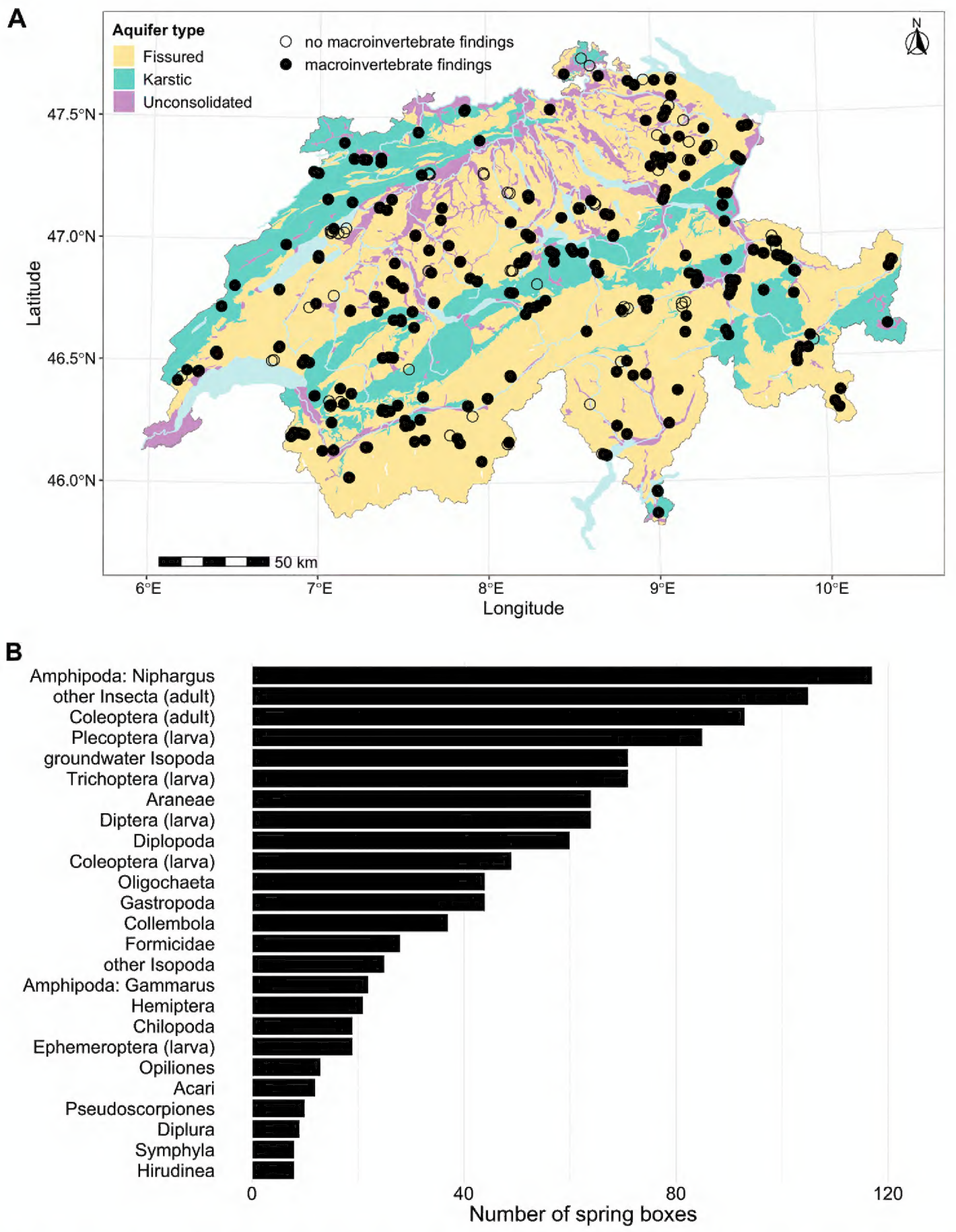

Figure 1. Sampling locations and macroinvertebrate occurrences **A** map of the main aquifer types in Switzerland (BAFU 2017) and the sampling sites (total n = 346). Filled circles indicate sites where macroinvertebrates were found and open circles mark sites where no macroinvertebrates were found. Geodata from the Swiss Federal Office of Topography **B** number of spring boxes (total n = 346) where each major macroinvertebrate group was found.

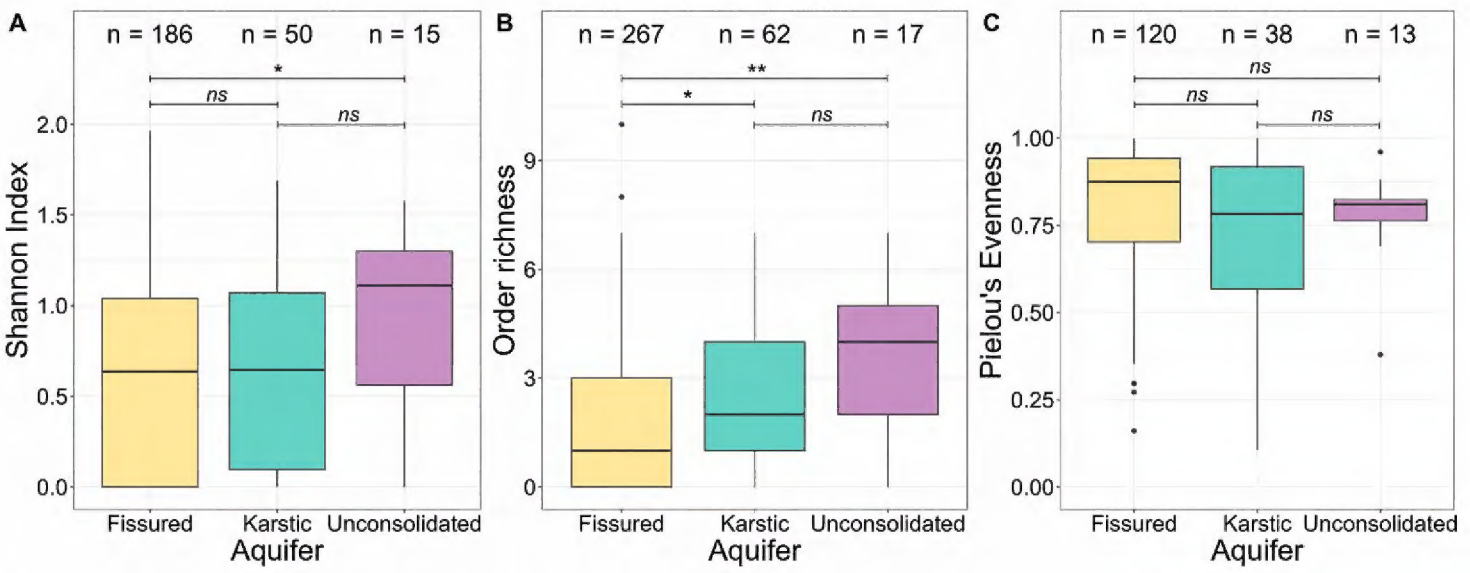

Figure 2. Local macroinvertebrate diversity at the order level per aquifer type **A** shannon Index (sites with at least one order included) **B** order richness (number of orders per site, all sites included), and **C** Pielou's Evenness (sites with at least two orders included). The thick horizontal lines show the median, the interior of each box represents the interquartile range (IQR) and the vertical lines represent minima and maxima, respectively (1.5 * IQR). The number of sites included for each analysis is shown on top of each boxplot, as well as the significance level between groups (pairwise Wilcoxon rank-sum test with Bonferroni correction, *ns* for $p > 0.05$, * for $p \leq 0.05$, ** for $p \leq 0.01$, *** for $p \leq 0.001$).

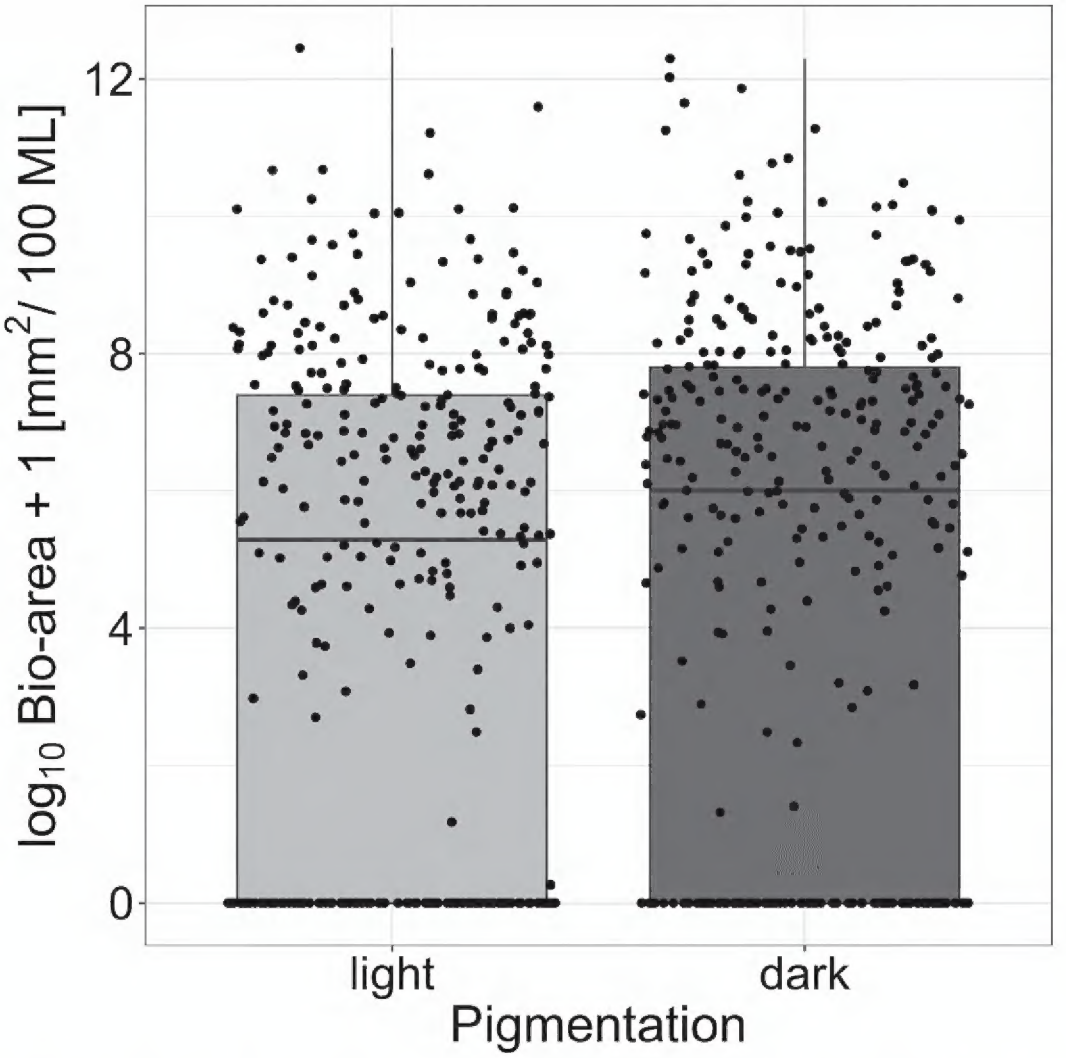

Figure 3. Standardized macroinvertebrate bio-area per site and pigmentation. Bio-area in mm^2 per 100 ML discharged groundwater and at $log_{10}(y+1)$-scale. The thick horizontal lines show the median, the interior of each box represents the interquartile range (IQR) and the vertical lines represent minima and maxima, respectively (1.5 * IQR).

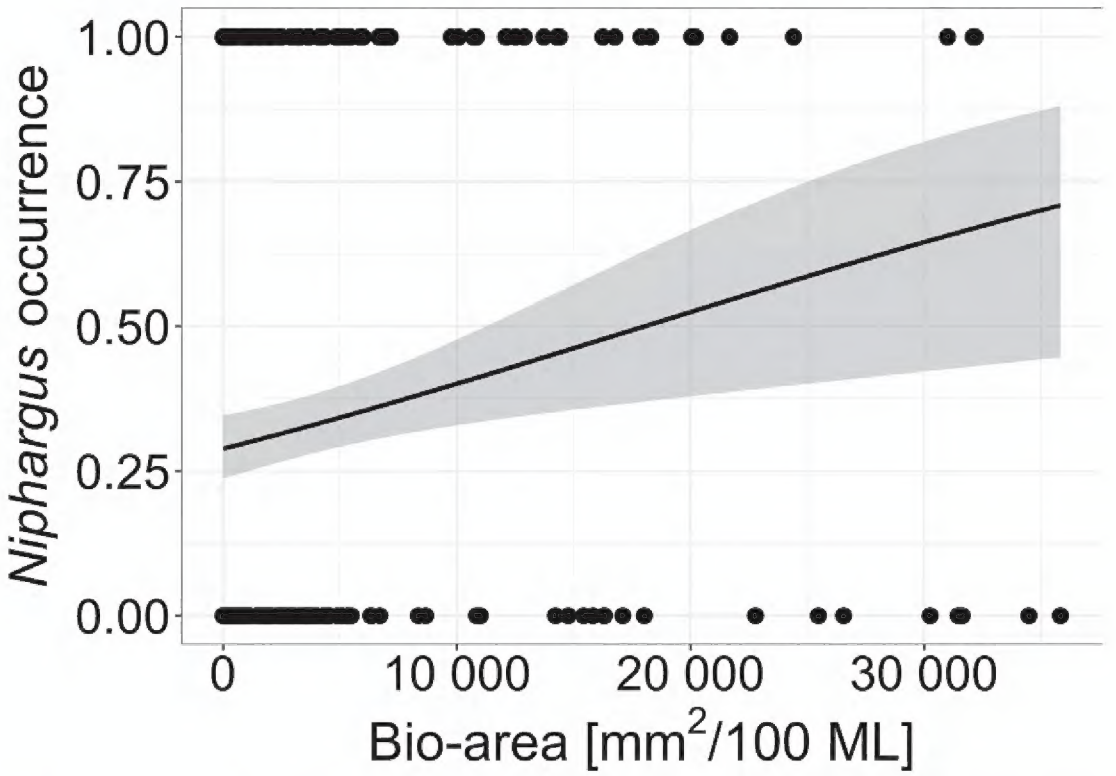

Figure 4. *Niphargus* occurrence in relation to the standardized bio-area of other macroinvertebrates. Predictions of binomial GLM are plotted as solid line with 95% confidence interval (shaded area). The binomial GLM was fitted to 334 sites with bio-area < 40,000 mm²/100 ML discharged groundwater. Bio-area in mm² per 100 ML discharged groundwater.

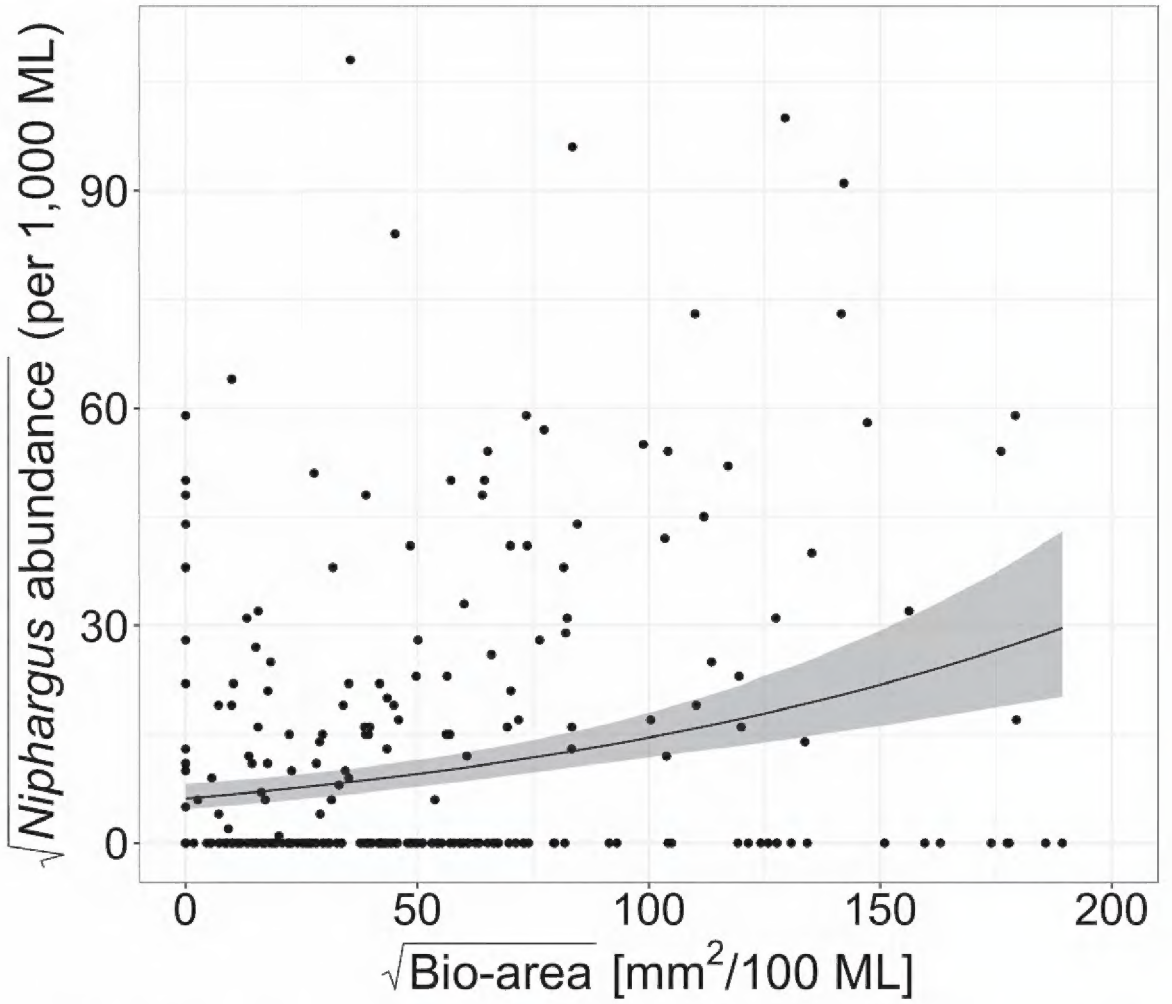

Figure 5. Fit of the ZINB model for *Niphargus* abundance and the standardized bio-area of other macroinvertebrates. Both variables were square-root-transformed. Bio-area in mm² per 100 ML discharged groundwater and *Niphargus* abundance per 1,000 ML discharged groundwater. For visualization, only points with values below 200 (x-axis) and 110 (y-axis) are plotted. The 95% confidence intervals are plotted in grey.

Discussion

Here, we provide a first countrywide overview of major groups of macroinvertebrates found in Swiss groundwater systems and address possible associations between groundwater and surface ecosystems through the assessment of these organisms. While groundwater amphipods are relatively well-known for this area (Altermatt et al. 2014, 2019; Fišer et al. 2017, 2018; Alther et al. 2021), the remaining groups have been understudied even at coarse taxonomic scale, hitherto prohibiting a first overview and understanding of their abundance and occurrence. Thus, we herewith contribute basic knowledge needed to successfully protecting and conserving the biodiversity of groundwater ecosystems (Wynne et al. 2021; Borko et al. 2022).

The application of a citizen science approach proved suitable to collect a broad range of macroinvertebrates from shallow groundwater aquifers. Collaborating with local drinking water providers to receive samples from spring boxes enabled us to obtain macroinvertebrates across a large geographic area, and from sampling sites that are otherwise not accessible to the public. In addition, the provided documentation on sampling duration and water discharge allowed highly standardized analyses of the samples. As such, it might be a suitable method to overcome the Racovitzan impediment (Ficetola et al. 2019), by providing large-scale, systematic data on groundwater fauna.

In line with previous studies (e.g., Hahn 2006; Hahn and Fuchs 2009; Johns et al. 2015), we found a high occurrence of macroinvertebrates in Swiss groundwater samples. The two stygobiotic taxa *Niphargus* and groundwater Isopoda (Asellidae, cf. *Proasellus*, unpigmented) had some of the highest absolute abundances. This pattern of high abundances of crustaceans is characteristic for groundwater systems (e.g., Sket 1999; Gibert and Deharveng 2002; Deharveng et al. 2009; Gibert and Culver 2009). Generally, we could associate the obtained macroinvertebrates with a combination of surface and subterranean, as well as terrestrial and aquatic ecosystems (i.e., groundwater, soil, and surface freshwater).

The high abundances and widespread occurrences of macroinvertebrates from surface waters such as Plecoptera, Trichoptera, and Ephemeroptera larvae (EPT taxa), *Gammarus*, as well as groups containing aquatic organisms (e.g., Diptera and Coleoptera larvae) were surprising and could reflect surface water infiltration and the interactions between above- and below-ground ecosystems (Griebler et al. 2010; Stein et al. 2010; Sinreich et al. 2012; Durkota et al. 2019). This result might be explained by the shallow depth from which most pipes drained the groundwater from the aquifers in our study. While some organisms might have been washed accidentally into the groundwater, some of these macroinvertebrate groups likely inhabit subterranean waters occasionally, for example during their larval stages (Stanford and Ward 1993; Malison et al. 2020). Consequently, they influence processes that occur in this ecotone, such as the transport of resources between surface and groundwater systems (Ward et al. 1998; Boulton 2000; Boulton et al. 2008; Barzaghi et al. 2017). Additionally, it has been suggested that some macroinvertebrates from surface waters migrate to the hyporheic zone or even deeper to escape disturbances such as droughts, floods, and pollution (Ward et al. 1998; Boulton 2000; Griebler and Avramov 2015; Durkota et al. 2019).

Local macroinvertebrate diversity and richness at the order level was significantly associated to the aquifer type. Unconsolidated aquifers had higher medians for diversity and richness compared to fissured aquifers. Similar observations have been made for example by Malard et al. (2009), who found that species richness of stygobiotic crustaceans was on average higher in porous aquifers. Previous studies also proposed that local groundwater faunal abundance and diversity is particularly influenced by the hydrological connectivity (e.g., Hahn 2006; Griebler et al. 2010; Foulquier et al. 2011). Therefore, a possible explanation for the observed higher diversity and richness in unconsolidated aquifers could be the fact that unconsolidated aquifers in Switzerland are found mainly along the main rivers of the Swiss Plateau, leading to higher surface water infiltration.

Aquifers are environments that lack photosynthetic primary production, and groundwater fauna relies largely on allochthonous energy sources imported from the surface (Gibert et al. 1994; Humphreys 2006; Foulquier et al. 2011), including organic matter inflows from plant materials, but also the immigration/inflow of invertebrates that can be predated on by groundwater organisms. An essential parameter for studying the transfer of energy between ecosystems is the biomass of macroinvertebrates (e.g., Machuca-Sepúlveda et al. 2022). In this study, we used the pigmentation of the macroinvertebrates as an approximate classification of hypogean (light-pigmented) and epigean fauna (dark-pigmented). This approach allowed us to overcome certain challenges of classifying macroinvertebrates into hypogean and epigean, as it did not depend on detailed taxonomic identification of the organisms. However, we acknowledge that pigmentation can exhibit a range of variations and thus might only give insights into the organism's affinity to hypogean or epigean ecosystems to a certain extent.

Of all the standardized macroinvertebrate bio-area obtained, 40% was classified as light-pigmented, approximating the hypogean fauna (including stygobiotic and terrestrial subterranean organisms).This portion is in accordance with the expected low biomass of groundwater ecosystems based on the limited availability of resources (Hose et al. 2022). Contrastingly, 60% of the standardized macroinvertebrate bio-area obtained was classified as dark-pigmented, which might approximate the amount of epigean, non-stygobiotic fauna found in our samples. Possibly, the high proportion of dark-pigmented macroinvertebrates could indicate a substantial input of allochthonous energy into groundwater ecosystems, including detritus and living organisms. The presence of non-stygobiotic organisms in shallow groundwater aquifers might also raise potential for predator-prey interactions among organisms. Yet, the degree and direction of trophic interactions between stygobiotic and non-stygobiotic organisms are not completely resolved and non-stygobiotic organisms could function as either prey or predators in groundwater ecosystems (Gibert et al. 1994). Since evidence suggests that some epigean species may use hypogean environments to escape unfavorable surface conditions, climate change might lead to altered biotic interactions between epigean and hypogean species (Vaccarelli et al. 2023).

We found a positive correlation between the macroinvertebrate bio-area and the occurrence and abundance of groundwater amphipods (*Niphargus* spp.). This could be due to local small-scale differences causing more organisms being washed out of the aquifers

into the pipes at some sites. For example, differences in the porosity of the groundwater systems or differences in the construction of the water drainage infrastructure, such as pipe size or depth from which the pipes drain the aquifers might lead to a higher rate of organisms being washed out at spring boxes (see Korbel et al. 2017 for a similar discussion). However, we did not find any significant difference in the obtained amount of standardized bio-area between aquifer types, which might disprove a possible effect of aquifer porosity. Alternatively, it could be an additional indication of the linkage between ecosystems, where a higher connectivity between the aquifers and the surface (and thus higher energy inputs from surface to groundwater environments) might correlate with a higher abundance of groundwater amphipods (e.g., Venarsky et al. 2018; Venarsky et al. 2023). Our study provides first data on the possibly tight linkage between hypogean and epigean ecosystems. We are aware that the lack of detailed taxonomic identification of the organisms and missing local environmental data limit the scope of further conclusions. Additional studies could reveal further insights into food web dynamics of groundwater ecosystems, for example through stable isotope analysis (Gibert et al. 1994; Saccò et al. 2019). Also, more data on groundwater quality could help to investigate the role of *Niphargus*, *Proasellus*, and other stygobites as bioindicators for groundwater monitoring.

Conclusion

Using citizen science samples collected by local drinking water providers, we identified major taxonomic groups of macroinvertebrates in shallow aquifers of Switzerland. Apart from obligate groundwater taxa, such as groundwater amphipods and isopods, we detected a substantial amount of macroinvertebrates associated with other surface and subsurface ecosystems. We also found a positive correlation between the macroinvertebrate biomass and the occurrence and abundance of groundwater amphipods, indicating a linkage between groundwater and other ecosystems. In particular, shallow aquifers might promote hydrological connectivity between surface water and groundwater. A better understanding of this linkage could help to conserve and manage groundwater ecosystems, especially as anthropogenic effects on surface ecosystems will affect groundwater ecosystems too.

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Supplementary material 1

Bio-area and biomass relationship
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Supplementary material 2

Detailed procedure for macroinvertebrate bio-area measurements
Authors: Ana Sofia Schneider, Mara Knüsel, Florian Altermatt
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Supplementary material 3

Supplementary results
Authors: Ana Sofia Schneider, Mara Knüsel, Florian Altermatt
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